Calcium from Diet and Supplements is Associated With Reduced Risk of Colorectal Cancer in a Prospective Cohort of Women

Andrew Flood,[1] Ulrike Peters,[2] Nilanjan Chatterjee,[2] James V. Lacey Jr.,[2] Catherine Schairer,[2] and Arthur Schatzkin[2]

[1]Division of Epidemiology, University of Minnesota, Minneapolis, Minnesota and [2]Division of Cancer Epidemiology and Genetics, National Cancer Institute, Bethesda, Maryland

Abstract

We investigated the association between calcium intake and colorectal cancer in a prospective cohort of 45,354 women without a history of colorectal cancer who successfully completed a 62-item National Cancer Institute/Block food-frequency questionnaire. Women were followed for an average of 8.5 years, during which time 482 subjects developed colorectal cancer. We used Cox proportional hazards models, with age as the underlying time metric, to estimate risk of colorectal cancer. Cut points between quintiles of energy-adjusted dietary calcium were 412, 529, 656, and 831 mg/day. We created categories for calcium from supplements as follows: 0 mg/day (n = 25,441), 0 to 400 mg/day (n = 9,452), 401 to 800 mg/day (n = 4,176), and >800 mg/day (n =6,285). Risk ratios and confidence intervals (95% CI) for increasing quintiles of dietary calcium relative to the lowest quintile were 0.79 (0.60-1.04), 0.77 (0.59-1.02), 0.78 (0.60-1.03), and 0.74 (0.56-0.98), P_{trend} = 0.05. For increasing categories of calcium from supplements, the risk ratios (and 95% CI) relative to no supplement use were 1.08 (0.87-1.34), 0.96 (0.70-1.32), and 0.76 (0.56-1.02), P_{trend} = 0.09. Simultaneously high consumption of calcium from diet and calcium from supplements resulted in even further risk reduction, RR = 0.54 (95% CI, 0.37-0.79) compared with low consumption of both sources of calcium. These data indicate that a difference of < 400 to > 800 mg of calcium per day was associated with an approximately 25% reduction in risk of colorectal cancer, and this reduction in risk occurred regardless of the source of the calcium (i.e., diet or supplements). (Cancer Epidemiol Biomarkers Prev 2005;14(1):126–32)

Introduction

Calcium has the potential to reduce risk of colorectal cancer through a variety of plausible biological mechanisms. Chief among these is the hypothesized ability of calcium to reduce the proliferative effect of secondary bile acids in the colon (1). Secondary bile acids, produced during the digestion of fat, are highly irritating to the epithelial cells of the colon, but calcium forms insoluble soaps with these bile acids thus neutralizing their ability to irritate the epithelial surface of the colon and thereby induce an increase in proliferation rates. Without the increase in cellular proliferation rates, the likelihood of individual initiated cells progressing to neoplasia or cancer would presumably be diminished. Alternatively, calcium may act through pathways independent of its ability to bind secondary bile acids and seemingly therefore to diminish proliferation rates. For example, calcium might also have direct effects on differentiation and apoptosis possibly related to the action of vitamin D, intracellular release of calcium, calmodulin activation, and subsequent phosphorylation of other cellular enzymes and activation of other signaling pathways (2).

Despite the biological plausibility of a calcium effect, earlier reviews and metaanalyses of the epidemiologic literature concluded that calcium intake does not have a significant effect on reduction of risk for colorectal cancer, or perhaps might only have a weak effect (3-5). Recent findings, however, have provided evidence to suggest that calcium does in fact have an important inverse association with colorectal neoplasia. Among these were two observational studies of adenoma recurrence, each showing reductions in risk for those with high intakes of calcium at baseline (6, 7) and eight reports from prospective cohort studies of calcium intake and incident colorectal cancer, all indicating reduced risk with higher calcium intake, although not always with statistical significance for all subanalyses (8-15). Additionally, in two separate double-blind, placebo-controlled clinical trials of calcium supplements, investigators found reductions in rates of adenoma recurrence (by 15% and 34% compared with placebo group) over 3 to 4 years of follow-up in patients with a history of adenomas (16, 17). In a third clinical trial of a combination supplement containing calcium and a variety of antioxidants, there was a similar 15% reduction in patients with recurrent adenomas in the intervention group compared with controls (18). Taken together, these recent studies suggest that the earlier reviews may have been premature in discounting the place that calcium might have in influencing risk of colorectal cancer.

In this paper, we present data from a prospective study of diet and colorectal cancer in a cohort of women selected from participants in a breast cancer-screening program. These analyses include specific consideration of calcium source (diet versus supplement) as well as anatomic subsite (colon versus rectum and distal colon versus proximal colon).

Materials and Methods

Study Population. The Breast Cancer Detection Demonstration Project (BCDDP) was a breast cancer-screening program conducted under the joint sponsorship of the National Cancer Institute and the American Cancer Society. The project ran from 1973 through 1980 and enrolled 283,222 women at 29 screening centers in 27 cities across the United States. In 1979, the National Cancer Institute established a follow-up cohort

Received 3/16/04; revised 7/26/04; accepted 8/5/04.

The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked advertisement in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

Requests for reprints: Andrew Flood, Division of Epidemiology, University of Minnesota, 1300 South Second Street, Suite 300, Minneapolis, MN 55454. Phone: 612-624-2891; Fax: 612-624-0315. E-mail: flood@epi.umn.edu

from a subset of the women who had participated in the BCDDP based on their breast cancer-screening status. The follow-up cohort included all 4,275 women from the screening program who had been diagnosed with breast cancer, all 25,114 women who had been diagnosed with benign breast disease, and all 9,628 who had been recommended for biopsy or breast surgery but did not have a surgical procedure (excluding women with a history of breast disease from the analysis did not materially affect the risk estimates, data not shown). An additional 25,165 women with no history of breast disease were matched with the above-listed subjects on age, time of entry into the screening program, ethnicity, screening center, and length of participation in the BCDDP for a total of 64,182 women selected for entry into the follow-up cohort. Of that number, 61,431 women (96%) completed the baseline questionnaire (given between 1979 and 1981) and were therefore eligible for further participation in the study. The Institutional Review Board of the National Cancer Institute approved the study, and all subjects provided written informed consent at the time of enrollment.

Participants subsequently completed a mailed questionnaire during three separate follow-up periods: 1987 to 1989, 1992 to 1995, and 1995 to 1998. Nonresponders to the questionnaires received vigorous follow-up including repeated mailings and phone calls.

For the purposes of the current analysis, entry into the analytic cohort took place at the time of the dietary assessment (1987-1989). We excluded from the study women who did not complete a questionnaire at that stage (n = 9,738), women with a diagnosis of colorectal cancer at the 1987 to 1989 questionnaire or earlier (n = 479), women whose reported entry date occurred after their exit date (n = 6, see definition of exit dates below), and women who skipped more than 30 items on their food frequency questionnaires or who had a reported total energy intake > 3,800 or < 400 kcal/day (n = 5,647). For this study, we also excluded 207 women with implausibly high intakes of calcium (reported level of consumption exceeding 3,000 mg/day), leaving 45,354 women in the final analytic cohort. Including women reporting consumption of more than 3,000 mg/day in the analyses did not materially alter the results (data not shown).

Cohort Follow-up. Follow-up began with completion of the dietary questionnaire (1987-1989). We defined ''end of study date'' as the date the subject completed the 1995 to 1998 questionnaire, or if the subject did not complete a 1995 to 1998 questionnaire, as the date of last contact in the 1995 to 1998 follow-up period. For participants not known to be deceased and with whom we had no contact in the 1995 to 1998 follow-up period, we imputed an end of study date by estimating the date on which subjects would have completed the 1995 to 1998 questionnaire (using mean time intervals from the rest of the cohort) had they actually completed one. We defined exit date from the study as the earliest among end of study date, date of colorectal cancer diagnosis, or date of death from cause other than colorectal cancer.

In the final analytic cohort, 90.3% (40,946 women) had complete follow-up through 1995 to 1998, meaning their exit date corresponded to either the date of their first colorectal cancer diagnosis, the date they filled out the 1995 to 1998 questionnaire, or their date of death from a cause other than colorectal cancer.

Case Ascertainment. We identified colorectal cancer cases from self-reports on the 1992 to 1995 and 1995 to 1998 questionnaires, from statewide cancer registries, and from the National Death Index (through 1997). We obtained pathology reports for 244 (79%) of the 309 women who provided self-reports of a diagnosis of colorectal cancer. The pathology reports confirmed 229 (94%) of the cases as adenocarcinoma of the colon or rectum (ICD-0 site codes 153.0-153.4 and 153.6-153.9 for colon cancer and 154.0-154.1 for rectal cancer). Because of this high correspondence between the self-reports and medical records, we included as cases the remaining 65 self-reports of colorectal cancer without pathology reports. Exclusion of these 65 cases did not materially affect the results (data not shown). Women with pathology reports contradicting self-reported colorectal cancers were not included as cases, unless they also appeared in a state cancer registry as described below. Pathology reports obtained for self-reported conditions unrelated to colorectal cancer identified 16 more cases of colorectal cancer. A search of the National Death Index identified an additional 107 individuals with death certificates indicating a diagnosis of colorectal cancer. Finally, we used last-known place of residence for each subject to match against state cancer registries for those states whose registries consented to participate in the study (accounting for 73.5% of the analytic cohort). Subjects residing in states with participating registries did not differ in any material way with respect to distribution of risk factors from subjects residing in states whose registries did not consent to participate. This procedure resulted in the identification of a further 65 colorectal cancer cases. Thus, the total number of cases in the analytic cohort over the follow-up period was 482.

Dietary Assessment. With the 1987 to 1989 questionnaire, respondents completed a 62-item National Cancer Institute/Block food frequency questionnaire to assess usual dietary intake over the previous year. Detailed descriptions of this food frequency questionnaire and its validity have appeared elsewhere (19-21). Software designed for this questionnaire yielded estimates of daily intakes for total energy, macronutrients, and micronutrients including calcium (21). A separate series of questions in the 1987 to 1989 questionnaire assessed intake of calcium from supplements, either multivitamin-type or calcium-specific.

Statistical Analysis. We used Cox proportional hazards regression (PROC PHREG in SAS version 6.12) with age as the underlying time metric to generate risk ratios and 95% confidence intervals (CI) for calcium from diet, calcium from supplements, and total calcium (the sum of dietary and supplemental calcium). All P values were two-sided. To test for trend, we entered calcium into the model as a continuous term.

We derived energy-adjusted dietary values using the residual method (for example, the mean value for dietary calcium in the analytic cohort added to the residual of dietary calcium regressed on total energy for each individual) as described by Willett (22). In analyzing calcium from supplements, because the distribution of intake was highly skewed (over half the cohort reported no calcium from supplements) we created four categories based on fixed levels of intake rather than dividing the cohort into quantiles. The four categories were 0 mg/day of calcium from supplements (n = 25,441), 1 to 400 mg/day (n = 9,452), 401 to 800 mg/day (n = 4,176), and > 800 mg/day (n = 6,285). For calcium from supplements and for total calcium, we did not adjust the energy intake of calcium from supplements because intake of a nutrient from supplement use is not fundamentally related to energy intake as would be a nutrient derived from food. Thus, in order to arrive at a single value for total calcium, we added the energy-adjusted dietary calcium to the raw value for intake of calcium from supplements. Using alternative energy adjustment methods such as the multivariate nutrient density method, the standard method (including total energy as a covariate), or the partition method for any of the analyses, with each offering a potentially unique interpretive perspective (23-25), did not materially affect the final results.

We also considered additional variables for inclusion into our models as potential confounders. In evaluating these risk factors, we entered each separately (by quintiles for continuous variables) into the energy-adjusted models for dietary and total calcium and the model for calcium from supplements. We judged a change of > 10% in the risk estimate for the highest quintile/category of calcium intake compared with the lowest from the age and energy-adjusted only model as evidence for confounding. We tested the following variables in this manner: smoking (ever/never), history of menopausal hormone replacement therapy (yes/no), education (through high school/more than high school), body mass index (kg/m^2), height, weekday physical activity index expressed in units of Metabolic Equivalent Time (26), alcohol, folate, vitamin D, fiber, meat, fat, fruits, vegetables, grains, and nonsteroidal antiinflammatory (NSAID) drug use (yes/no). NSAIDs included aspirin, ibuprofen (Advil, Motrin, Nuprin), Naprosyn, and other pain-relieving drugs, but excluded Tylenol. We defined women to be users of NSAIDs if they had used these drugs at least once a week for at least 1 year. After performing all of these tests, we found that none of the factors listed above generated any material changes in either the dietary calcium model, the total calcium model, or the supplemental calcium model (data not shown), and thus did not include them in any of the final calcium models. Likewise, adjusting for reported colorectal cancer-screening or restricting the cohort to only those subjects who had no prior history of breast disease resulted in no qualitative differences in risk estimates. Simultaneous inclusion of all these potential confounders in the proportional hazards regression models also did not produce any material changes in the results from those obtained in the age- and energy-adjusted models.

Results

Women in the BCDDP follow-up cohort completed the dietary questionnaire at an average of 61.9 years of age and contributed an average of 8.5 years of follow-up. Table 1 presents baseline characteristics of the analytic cohort at the time of entry into the study according to quintiles of dietary calcium intake. These baseline characteristics were generally similar across quintiles of dietary calcium intakes with only relatively modest increases or decreases in a few food groups or nutrients (e.g., red meat, percentage of energy from fat, alcohol, dietary folate, and folate from supplements). The only exception was dietary vitamin D which showed a greater than 4-fold increase across quintiles of dietary calcium. The strong correlation between dietary calcium and vitamin D will receive further attention below. The baseline characteristics in categories of supplemental calcium and quintiles of total calcium had a very similar pattern (data not shown) with the only exception being that NSAID use was slightly more common in the high category of intake compared with the low (43.5% versus 36.0%).

In the first quintile of dietary calcium intake, energy-adjusted median consumption was 337 mg/day, and in the fifth quintile it was 985 mg/day yielding a range of intake just under 650 mg/day. Across quintiles of dietary calcium, we observed a reduction in risk for colorectal cancer with increasing intake (Table 2). Quintile 5 had a relative risk (RR) of 0.74 (95% CI, 0.56-0.98) compared with quintile 1 (P for trend = 0.05). Across categories of calcium intake from supplements, we had a somewhat broader range of intake (0 mg/day in the low category compared with 1,130 mg/day median in the high category), and we saw a similar association with colorectal cancer in the high compared with the low category (RR = 0.76, 95% CI, 0.56-1.02; P for trend = 0.09). Calcium in the diet and calcium from supplements were uncorrelated in this cohort (r = 0.01) indicating that the association of colorectal cancer with calcium from one source was completely independent of the association with calcium from the other.

To determine if there was additional benefit from having high intakes of both dietary calcium and calcium from supplements, we did a secondary analysis in which we combined categories of dietary and supplemental calcium intake and entered these into a single proportional hazards regression model (Table 3). We combined categories with similar main effects (dietary calcium quintiles 2-5 and supplemental calcium categories 1-3) into a single category for both dietary and supplemental calcium. Women who were simultaneously in the highest category of intake from both sources of calcium had even further reduction in risk

Table 1. Baseline characteristics of 45,354 women in the BCDDP follow-up cohort according to quintile of dietary calcium intake (all values are percentages or means in units listed)

	Quintile of dietary calcium intake*				
	1	2	3	4	5
	<412.3 (mg/d)*	412.4-528.9 (mg/d)*	529.0-656.2 (mg/d)*	656.3-830.9 (mg/d)*	≥830.9 (mg/d)*
Calcium—diet (mg)*	332.1	471.6	589.8	736.6	1,057.7
Calcium—supplements (mg)	286.4	322.3	322.1	306.7	391.8
Dietary vitamin D (IU)*	66.3	105.2	133.5	175.2	270.8
Age	61.1	61.7	61.9	62.2	62.4
Energy (kcal)	1,273	1,258	1,283	1,288	1,265
Body mass index (kg/m^2)	24.7	24.7	24.7	24.7	24.5
Height (inches)	63.8	63.9	64.0	64.0	64.1
Physical activity (metabolic equivalent time)	56.6	56.9	57.2	57.2	56.7
Alcohol (g)	5.6	4.3	3.9	3.6	2.8
Vegetables (servings)*	2.6	2.9	3.0	3.0	3.0
Fruit (servings)*	1.2	1.3	1.3	1.3	1.3
Red meat (g)*	39.1	34.8	32.3	29.7	24.0
Energy from fat (%)	37.6	35.8	35.5	34.4	31.4
Fiber (g)*	9.3	11.2	11.4	11.6	11.6
Folate from diet (μg)*	203	254	266	274	285
Supplemental folate (μg)	139	151	155	166	177
NSAID users (%)	37.8	37.3	38.5	40.0	39.9
Smokers, current/former (%)	45.3	43.4	42.7	42.7	41.6
More than high school education (%)	40.6	43.9	45.7	48.4	52.6

*Adjusted for total energy using the residual method as described in the text.

Table 2. Age-adjusted relative risk of colorectal cancer by quintile of dietary calcium, category of calcium from supplements, and quintile of total calcium (dietary calcium plus calcium from supplements)

	Quintile or category (for supplements) of calcium intake					
	Q1	Q2	Q3	Q4	Q5	P_{trend}
Dietary calcium*						
Median intake (range), mg	337 (<412)	473 (412-528)	588 (529-655)	733 (656-830)	985 (>830)	
Cases	113	93	92	95	91	
RR (95% CI)	1.00 (reference)	0.79 (0.60-1.04)	0.77 (0.59-1.02)	0.78 (0.60-1.03)	0.74 (0.56-0.98)	0.05
Calcium from supplements						
Median intake (range), mg	0 (0)	130 (1-400)	540 (401-800)	1,130 (>800)		
n	25,441	9,452	4,176	6,285		
Cases	273	115	44	52		
RR (95% CI)	1.00 (reference)	1.08 (0.87-1.34)	0.96 (0.70-1.32)	0.76 (0.56-0.98)		0.09
Total calcium†						
Median intake (range), mg	377 (<472)	551 (472-635)	728 (636-844)	1,003 (845-1,270)	1,676 (>1,270)	
Cases	102	110	86	106	80	
RR (95% CI)	1.00 (reference)	1.03 (0.79-1.35)	0.80 (0.60-1.06)	0.96 (0.73-1.26)	0.74 (0.55-0.99)	0.02

*Based on residual method for energy adjustment of dietary calcium intake.

compared with those who were in the low categories on each (RR = 0.54, 95% CI, 0.37-0.79). Women who were in the high-intake quintiles of dietary calcium but in the low category for calcium from supplements had intermediate levels of risk reduction (RR = 0.82, 95% CI, 0.65-1.02). The grouping with high intake of calcium from supplements but low intake from diet showed no reduction in risk (RR = 1.05), but this cell was sparsely populated (n = 1,209), and the confidence intervals were wide (95% CI, 0.62-1.77).

We had information on subsite specificity for 358 of the 482 cases of colorectal cancer. Using this information, we observed a slightly stronger and more consistent association between calcium intake and cancers of the colon than we did with cancers of the rectum (Table 4). The number of cases of rectal cancer, however, was small (n = 74), and the confidence intervals were therefore wide, making it difficult to differentiate between the risk estimates for the two sites (colon versus rectum). When analyzing the association between either dietary or total calcium with cancer in the distal colon (defined as distal to the splenic flexure but not including rectum, n = 112 cases) or in the proximal colon (n = 172 cases), we observed no qualitative differences from the associations we observed using colon cancer in general as the case definition.

Vitamin D mediates the absorption of calcium in the small intestine, and therefore consideration of vitamin D status could provide additional insight into the role of calcium in reducing risk of colorectal cancer. Unfortunately, we had no direct measure of sun exposure, a major source of circulating vitamin D. Thus, our ability to measure vitamin D status and correctly classify subjects on this exposure was necessarily limited. Also, in the BCDDP cohort, and in the U.S. diet generally, vitamin D and calcium both come primarily from milk and dairy products (five dairy foods contributed over 90% of the dietary calcium in the BCDDP cohort). Thus, dietary calcium and dietary vitamin D are highly correlated (r = 0.87 in the BCDDP cohort) making it difficult to disentangle their independent associations with colorectal cancer. Correlations with total vitamin D intake (from both diet and supplements) were substantially lower (r = 0.33 for dietary calcium and 0.10 for calcium from supplements) making it more feasible to assess the effects of total vitamin D intake on the association of calcium with colorectal cancer. We did an analysis of calcium and colorectal cancer after stratifying total vitamin D intake, but these did not reveal statistical effect modifications of the calcium association by vitamin D intake from diet and supplements (data not shown).

Discussion

In our prospective analyses of calcium from diet and calcium from supplements, we observed an approximately 25% decrease in risk with higher intakes of each and even higher reduction in risk, roughly 45%, with simultaneously high consumption of both. These associations were generally not subsite-dependent, nor did they change after adjustment for a wide variety of potential confounders or stratification on body mass index, NSAID use, smoking status, or history of prior cancer other than colorectal cancer (data not shown). Likewise, the results were not affected by the modification of various criteria we established for inclusion of subjects in the analyses, and although we cannot rule out the possibility that the calcium-colorectal cancer association we observed could have been different among the women excluded for providing inadequate dietary information, there is no reason to believe that it would be.

It is especially notable that the risk reduction in the BCDDP cohort was present regardless of the source of calcium, that the sources of calcium were almost completely uncorrelated, and that simultaneously consuming high levels of calcium from both diet and supplements further reduced risk. These observations suggest that it was calcium per se, and not merely dairy foods or some other unmeasured confounding variable that we did not include in our analyses, that accounted for the reduction in risk.

Although we observed an inverse association regardless of the source of calcium, there did seem to be a difference between calcium from diet and calcium from supplements in the level at which we observed the risk reduction. For calcium from diet, intake of > 400 mg/day resulted in a marked reduction in risk, whereas in the case of calcium from supplements, we observed no association at an intake of < 800 mg/day. This discrepancy might be attributed to differences between true intake and self-reported estimates of intake for supplements and diet, or it may be the result of differences between the bioavailability of calcium from diet as compared with that from supplements.

Our results provide additional evidence that calcium intake is inversely related to colorectal cancer. Among the earlier reports (prior to 1997) only two prospective studies showed statistically significant, inverse associations with colorectal cancer or adenomas (27, 28), whereas eight others did not (29-35). Many of these studies were relatively small, however, and were thus underpowered to observe relative risks on the order of 0.75 to 0.85, the magnitude of risk reduction commonly observed in subsequent clinical trials and prospective studies.

In fact, six of the eight earlier studies did have nonsignificant risk estimates consistent with this range. As mentioned, since 1997, there have been eight additional prospective cohort studies (8-15), generally with greater power, and each of these reported inverse associations between calcium and colorectal cancer. In some cases, the associations were statistically significant only for subsets of the population or subclassifications of disease site [e.g., among women without a family history of colorectal cancer (9), for cancers of the colon but not the rectum (12), for distal rather than proximal colon (10), or among men but not women (10)]. In only one study, the NYU cohort, were there no statistically significant associations (8). Notwithstanding a relatively small study size (only 100 cases) in this cohort and thus wide confidence intervals, the relative risk associated with the high quartile of intake compared with the lowest was 0.71, consistent with the other studies. This body of literature from prior prospective studies together with the results from the present analyses and three clinical trials demonstrating the ability of calcium supplements to reduce adenomatous polyp recurrence (16-18), present a consistent picture of a modest but real reduction in risk, perhaps of 15% to 25%, with higher intakes of calcium (> 800-1,200 mg/day).

Of the 17 prospective studies that have considered calcium and colorectal neoplasia, only six (10, 12, 14, 15, 28, 34, 36) have undertaken subsite analyses. Of these, Wu et al. (10), Terry et al. (14), and Stemmermann et al. (28), the latter having very few cases, observed a more pronounced effect in the distal colon compared with the proximal colon, but only in the study by Wu et al. was the association more than marginal. By contrast, McCullough et al. (15) observed a stronger effect not in the distal but in the proximal colon, but only for total calcium and not dietary calcium or milk or dairy products. When comparing colon to rectum, Jarvinen et al. (12) and McCullough et al. both observed a difference (with the stronger effect in the colon), whereas Kampman et al. (34) and Terry et al. (14), saw no differences in risk estimates between these anatomic sites. In light of these few mixed findings, our results showing no substantial differences (or perhaps marginal ones) among subsites are consistent with the existing body of literature. Further studies are required before it will be possible to make more confident statements about the possible subsite specificity of the calcium effect, but at present, the evidence does not offer substantial support for any hypothesized differences in risk reduction from calcium intake between proximal and distal colon or between colon and rectum.

Our results, and the results from previous related studies, raise interesting questions with regard to the mechanism by which calcium may have the effect of reducing risk of colorectal cancer. If calcium acted by neutralizing secondary bile acids that would otherwise provide an irritative, proliferative stimulus to the epithelial lining of the colon, we would expect higher calcium concentration in the lumen to affect indicators of proliferation such that they would be decreased. Yet in 14 separate randomized clinical trials of calcium supplements or high-calcium (i.e., dairy) foods (37-50), 10 saw no change in proliferation rates in colorectal mucosal biopsies.

Of the four clinical trials that did see a reduction in proliferation rates, two were among familial adenomatous polyposis patients (47, 49) with the authors of one of these studies commenting that the effect was typically confined to individuals with high proliferation rates at baseline (47). In contrast to these results, however, Stern et al. (50) saw no notable changes in mucosal risk factors after a 9-month placebo-controlled trial of calcium supplementation in a similar population with familial polyposis. A third study saw a reduction in proliferation rates among patients with a family history of colorectal cancer, although without polyposis (49), and the final study showing a reduced proliferation rate with calcium supplementation saw the effect only in patients who received a dose of 2,000 mg/day (lower doses produced no effect; ref. 48). In one of the few studies that considered location of mitotic activity within the crypt, the authors observed that although calcium supplementation did not reduce overall proliferation rates, it did normalize the distribution of proliferating cells within the crypt (39). These latter observations do suggest that the effect of calcium may be subtler than merely reducing proliferation generally, and that it may act only at high doses (at least for calcium from supplements, consistent with our results), and that it may affect only subsets of the population. Furthermore, almost all of the proliferation studies relied on rectal biopsies (37, 39, 40, 42-44) and thus were unable to assess potentially different effects of calcium supplementation in the colon as distinct from the rectum. Nonetheless, the predominantly null results from these clinical trials do not provide strong evidence in favor of the hypothesis that calcium will reduce risk of colorectal cancer by downward influence on rates of proliferation in the epithelium.

Furthermore, proliferation rates themselves may not be indicative of future risk for colorectal neoplasia as described in a follow-up study by Sandler et al. (51) in which proliferation at baseline was, if anything, inversely correlated with risk of adenoma 3 years subsequently. Thus, although evidence accumulates that calcium has an inverse association with colorectal cancer, the evidence in support of the hypothesis that it does so by reducing the proliferative effects of secondary bile acids remains weak at best.

The presence of fat in the diet, because of its influence on the secretion of bile acids into the gut, should be an important risk factor for colorectal cancer if the hypothesized action of calcium to neutralize these bile acids in the lumen were to be truly related to risk of colorectal cancer. The epidemiological literature on fat and colorectal cancer, however, is not consistent (52). In particular, there was no association between fat and colorectal cancer in a separate analysis of the BCDDP cohort (53), and yet, we still observed

Table 3. Age-adjusted relative risk of colorectal cancer cross-classifying dietary calcium and calcium from supplements

	Calcium from diet	
	Q1	Q2-Q5
Calcium from supplements	Median intake, 337 mg (range, <412 mg)*	Median intake, 655 mg (range, ≥412 mg)*
Categories 1-3 Median intake, 130 mg (range, ≤800 mg)	1.00 (reference) n = 7,861 (97 cases)	0.82 (0.65-1.02) n = 31,208 (335 cases)
Category 4 Median intake, 1,270 mg (range, >800 mg)	1.05 (0.62-1.77) n = 1,209 (16 cases)	0.54 (0.37-0.79) n = 5,076 (36 cases)

NOTE: P value for interaction term for calcium from diet and calcium from supplements was not significant.
*Based on residual method for energy adjustment of dietary calcium intake.

Table 4. Age-adjusted relative risk of colorectal cancer by subsite for quintiles of dietary calcium and total calcium (dietary calcium plus calcium from supplements)

	Quintile of calcium intake					
	Q1	Q2	Q3	Q4	Q5	P_{trend}
Rectum (74 cases)						
Dietary calcium*						
Range of intake (mg)	<412	412-528	529-655	656-830	>830	
RR (95% CI)	1.00 (reference)	0.73 (0.35-1.55)	1.02 (0.51-2.01)	0.82 (0.40-1.69)	0.87 (0.43-1.77)	0.85
Total calcium†						
Range of intake (mg)	<472	472-635	636-844	845-1,270	>1,270	
RR (95% CI)	1.00 (reference)	1.19 (0.57-2.48)	1.10 (0.52-2.32)	1.23 (0.60-2.53)	0.93 (0.43-2.01)	0.30
Colon (284 cases)						
Dietary calcium*						
Range of intake (mg)	<412	412-528	529-655	656-830	>830	
RR (95% CI)	1.00 (reference)	0.78 (0.56-1.11)	0.66 (0.46-0.94)	0.70 (0.49-0.99)	0.62 (0.43-0.90)	0.02
Total calcium†						
Range of intake (mg)	<472	472-635	636-844	845-1,270	>1,270	
RR (95% CI)	1.00 (reference)	0.84 (0.59-1.18)	0.66 (0.46-0.96)	0.78 (0.55-1.11)	0.69 (0.48-0.99)	0.10
Distal colon (112 cases)						
Dietary calcium*						
Range of intake (mg)	<412	412-528	529-655	656-830	>830	
RR (95% CI)	1.00 (reference)	0.64 (0.36-1.14)	0.70 (0.40-1.23)	0.76 (0.44-1.31)	0.66 (0.37-1.16)	0.54
Total calcium†						
Range of intake (mg)	<472	472-635	636-844	845-1,270	>1,270	
RR (95% CI)	1.00 (reference)	0.76 (0.43-1.34)	0.86 (0.49-1.49)	0.71 (0.40-1.26)	0.71 (0.40-1.26)	0.26
Proximal colon (172 cases)						
Dietary calcium*						
Range of intake (mg)	<412	412-528	529-655	656-830	>830	
RR (95% CI)	1.00 (reference)	0.88 (0.57-1.35)	0.63 (0.39-1.01)	0.65 (0.41-1.04)	0.60 (0.38-0.97)	0.01
Total calcium†						
Range of intake (mg)	<472	472-635	636-844	845-1,270	>1,270	
RR (95% CI)	1.00 (reference)	0.89 (0.57-1.38)	0.54 (0.33-0.90)	0.83 (0.53-1.29)	0.68 (0.42-1.08)	0.24

*Based on residual method for energy adjustment of dietary calcium intake.
† Based on residual method for energy adjustment of dietary calcium intake plus unadjusted supplemental calcium intake.

a significant inverse association for calcium in this population. Again, these results are not consistent with neutralizing secondary bile acids as the primary mechanism by which calcium reduces risk of colorectal cancer.

Finally, we observed no difference in the calcium association across strata of dietary vitamin D, although it is important to recognize that dietary vitamin D is an imperfect surrogate for serum vitamin D because a large portion of circulating vitamin D derives from exposure to the sun, something for which we had no information in the BCDDP cohort. Other investigators have pointed out the decreasing local rates of colorectal cancer when moving from the north to the south and have postulated increased circulating vitamin D (a result of increased sun exposure) as a possible explanation (54). Given the high correlation between dietary calcium and dietary vitamin D (the primary sources of both were dairy foods), it is tempting to propose that the calcium effect we observed was simply the result of confounding by vitamin D. However, there was no correlation between dietary vitamin D and calcium from supplements, yet we still observed an inverse association between supplemental calcium and colorectal cancer. This does not exclude an important, independent role for vitamin D, but it does suggest that calcium in this case is not simply a surrogate for vitamin D intake.

If the ability of calcium in the colonic lumen to form inert soaps with secondary bile acids is not an explanation that satisfactorily accounts for the existing data on calcium and colorectal neoplasia, the alternative most likely involves a mechanism related to direct effects of calcium on the colonic epithelial cells. It is important to consider, however, that circulating levels of calcium are subject to tight homeostatic regulation and vary only within a narrow range. Therefore, whatever beneficial effect calcium may have on colorectal epithelial cells, it is not likely to be related to changes in serum concentration of the mineral. Lamprecht and Lipkin (2) summarize the cellular modes of action that can explain the effects of calcium in reducing risk of colorectal cancer through the activation of calcium-sensing receptors on the luminal surface of intestinal epithelial cells. These receptors are G protein–coupled receptors that activate diverse intracellular signaling pathways that can potentially influence epithelial cell differentiation. This mechanism could explain the ability of calcium to reduce the risk of colorectal neoplasia even though it has no effect on proliferation rates, although at the same time its serum concentration does not deviate from the narrow range of homeostatic control.

In summary, despite earlier reviews expressing some hesitation with respect to a judgment on calcium's beneficial effects, our study provides further evidence to support what is now an increasing body of literature indicating that calcium intake will reduce risk of colorectal cancer.

Acknowledgments

We thank Leslie Carroll at IMS, Inc., Silver Spring, MD for computer support and Cathy Ann Grundmayer, Susan Englehart, and the BCDDP staff of Westat, Inc., Rockville, MD for assistance in collecting and processing data for the study. The authors wish to acknowledge the California Department of Health Services, Cancer Surveillance Section; the Florida Cancer Data System under contract to the state Department of Health; the Maryland Cancer Registry, Maryland Department of Health and Mental Hygiene; the Michigan Cancer Surveillance Program within the Division for Vital Records and Health Statistics, Michigan Department of Community Health; the Division of Health Statistics, Pennsylvania Department of Health; the Tennessee Cancer Registry; the Texas Department of Health; and the states of Arizona, Georgia, Hawaii, Idaho, Iowa, New Jersey, New York, North Carolina, Ohio, Oregon, and Rhode Island for providing data from their cancer registries for use in these analyses.

References

1. Newmark HL, Wargovich MJ, Bruce WR. Colon cancer and dietary fat, phosphate, and calcium: a hypothesis. J Natl Cancer Inst 1984;72:1323–5.
2. Lamprecht SA, Lipkin M. Cellular mechanisms of calcium and vitamin D in the inhibition of colorectal carcinogenesis. Ann N Y Acad Sci 2001;952: 73–87.
3. Martinez ME, Willett WC. Calcium, vitamin D, and colorectal cancer: a review of the epidemiologic evidence. Cancer Epidemiol Biomarkers Prev 1998;7:163–8.
4. World Cancer Research Fund/American Institute for Cancer Research. Food, nutrition and the prevention of cancer: a global perspective. Washington, DC: American Institute for Cancer Research; 1997.
5. Bergsma-Kadijk JA, van't Veer P, Kampman E, Burema J. Calcium does not protect against colorectal neoplasia. Epidemiology 1996;7:590–7.
6. Hyman J, Baron JA, Dain BJ, et al. Dietary and supplemental calcium and the recurrence of colorectal adenomas. Cancer Epidemiol Biomarkers Prev 1998;7:291–5.
7. Martinez ME, Marshall JR, Sampliner R, Wilkinson J, Alberts DS. Calcium, vitamin D, and risk of adenoma recurrence (United States). Cancer Causes Control 2002;13:213–20.
8. Kato I, Akhmedkhanov A, Koenig K, Toniolo PG, Shore RE, Riboli E. Prospective study of diet and female colorectal cancer: the New York University Women's Health Study. Nutr Cancer 1997;28:276–81.
9. Sellers TA, Bazyk AE, Bostick RM, et al. Diet and risk of colon cancer in a large prospective study of older women: an analysis stratified on family history (Iowa, United States). Cancer Causes Control 1998; 9:357–67.
10. Wu K, Willett WC, Fuchs CS, Colditz GA, Giovannucci EL. Calcium intake and risk of colon cancer in women and men. J Natl Cancer Inst 2002; 94:437–46.
11. Pietinen P, Malila N, Virtanen M, et al. Diet and risk of colorectal cancer in a cohort of Finnish men. Cancer Causes Control 1999; 10:387–96.
12. Jarvinen R, Knekt P, Hakulinen T, Aromaa A. Prospective study on milk products, calcium and cancers of the colon and rectum. Eur J Clin Nutr 2001;55:1000–7.
13. Zheng W, Anderson KE, Kushi LH, et al. A prospective cohort study of intake of calcium, vitamin D, and other micronutrients in relation to incidence of rectal cancer among postmenopausal women. Cancer Epidemiol Biomarkers Prev 1998;7:221–5.
14. Terry P, Baron JA, Bergkvist L, Holmberg L, Wolk A. Dietary calcium and vitamin D intake and risk of colorectal cancer: a prospective cohort study in women. Nutr Cancer 2002;43:39–46.
15. McCullough ML, Robertson AS, Rodriguez C, et al. Calcium, vitamin D, dairy products, and risk of colorectal cancer in the cancer prevention study II nutrition cohort (United States). Cancer Causes Control 2003;14:1–12.
16. Baron JA, Beach M, Mandel JS, et al. Calcium supplements for the prevention of colorectal adenomas. Calcium Polyp Prevention Study Group. N Engl J Med 1999;340:101–7.
17. Bonithon-Kopp C, Kronborg O, Giacosa A, Rath U, Faivre J. Calcium and fibre supplementation in prevention of colorectal adenoma recurrence: a randomised intervention trial. European Cancer Prevention Organisation Study Group. Lancet 2000;356:1300–6.
18. Hofstad B, Almendingen K, Vatn M, et al. Growth and recurrence of colorectal polyps: a double-blind 3-year intervention with calcium and antioxidants. Digestion 1998;59:148–56.
19. Block G, Hartman AM, Dresser CM, Carroll MD, Gannon J, Gardner L. A data-based approach to diet questionnaire design and testing. Am J Epidemiol 1986;124:453–69.
20. Block G, Hartman AM, Naughton D. A reduced dietary questionnaire: Development and validation. Epidemiology 1990;1:58–64.
21. National Cancer Institute, Information Management Services I, Block Dietary Data Systems. DIETSYS version 3.0 user's guide. Bethesda, MD: National Cancer Institute; 1994.
22. Willett W. Nutritional Epidemiology. New York: Oxford University Press; 1990.
23. Willett WC, Howe GR, Kushi LH. Adjustment for total energy intake in epidemiologic studies. Am J Clin Nutr 1997;65:1220–8s.
24. Wacholder S, Schatzkin A, Freedman LS, Kipnis V, Hartman A, Brown CC. Can energy adjustment separate the effects of energy from those of specific macronutrients? Am J Epidemiol 1994;140:848–55.
25. Willett W, Stampfer M. Implications of total energy intake for epidemiologic analyses. 2nd ed. In: Willett W, editor. New York: Oxford University Press; Nutritional Epidemiology 1998;:273–301.
26. Ainsworth BE, Haskell WL, Whitt MC, et al. Compendium of physical activities: an update of activity codes and MET intensities. Med Sci Sports Exerc 2000;32:S498–516.
27. Garland C, Shekelle RB, Barrett-Connor E, Criqui MH, Rossof AH, Paul O. Dietary vitamin D and calcium and risk of colorectal cancer: a 19-year prospective study in men. Lancet 1985;1:307–9.
28. Stemmermann GN, Nomura A, Chyou PH. The influence of dairy and nondairy calcium on subsite large-bowel cancer risk. Dis Colon Rectum 1990;33:190–4.
29. Wu AH, Paganini-Hill A, Ross RK, Henderson BE. Alcohol, physical activity and other risk factors for colorectal cancer: a prospective study. Br J Cancer 1987;55:687–94.
30. Gaard M, Tretli S, Loken EB. Dietary factors and risk of colon cancer: a prospective study of 50,535 young Norwegian men and women. Eur J Cancer Prev 1996;5:445–54.
31. Slob IC, Lambregts JL, Schuit AJ, Kok FJ. Calcium intake and 28-year gastrointestinal cancer mortality in Dutch civil servants. Int J Cancer 1993;54:20–5.
32. Bostick RM, Potter JD, Sellers TA, McKenzie DR, Kushi LH, Folsom AR. Relation of calcium, vitamin D, and dairy food intake to incidence of colon cancer among older women. The Iowa Women's Health Study. Am J Epidemiol 1993;137:1302–17.
33. Martinez ME, Giovannucci EL, Colditz GA, et al. Calcium, vitamin D, and the occurrence of colorectal cancer among women. J Natl Cancer Inst 1996;88:1375–82.
34. Kampman E, Giovannucci E, van't Veer P, et al. Calcium, vitamin D, dairy foods, and the occurrence of colorectal adenomas among men and women in two prospective studies. Am J Epidemiol 1994;139:16–29.
35. Kampman E, Goldbohm RA, van den Brandt PA, van't Veer P. Fermented dairy products, calcium, and colorectal cancer in The Netherlands Cohort Study. Cancer Res 1994;54:3186–90.
36. Indar-Brown K, Noreberg C, Madar Z. Glycemic and insulinemic responses after ingestion of ethnic foods by NIDDM and healthy subjects. Am J Clin Nutr 1992;55:89–95.
37. Holt PR, Atillasoy EO, Gilman J, et al. Modulation of abnormal colonic epithelial cell proliferation and differentiation by low-fat dairy foods: a randomized controlled trial. JAMA 1998;280:1074–9.
38. Weisgerber UM, Boeing H, Owen RW, Waldherr R, Raedsch R, Wahrendorf J. Effect of long-term placebo-controlled calcium supplementation on sigmoidal cell proliferation in patients with sporadic adenomatous polyps. Gut 1996;38:396–402.
39. Bostick RM, Fosdick L, Wood JR, et al. Calcium and colorectal epithelial cell proliferation in sporadic adenoma patients: a randomized, double-blinded, placebo-controlled clinical trial. J Natl Cancer Inst 1995;87: 1307–15.
40. Bostick RM, Potter JD, Fosdick L, et al. Calcium and colorectal epithelial cell proliferation: a preliminary randomized, double-blinded, placebo-controlled clinical trial. J Natl Cancer Inst 1993;85:132–41.
41. Cascinu S, Ligi M, Del Ferro E, et al. Effects of calcium and vitamin supplementation on colon cell proliferation in colorectal cancer. Cancer Invest 2000;18:411–6.
42. Alberts DS, Einspahr J, Ritenbaugh C, et al. The effect of wheat bran fiber and calcium supplementation on rectal mucosal proliferation rates in patients with resected adenomatous colorectal polyps. Cancer Epidemiol Biomarkers Prev 1997;6:161–9.
43. Bostick RM, Boldt M, Darif M, Wood JR, Overn P, Potter JD. Calcium and colorectal epithelial cell proliferation in ulcerative colitis. Cancer Epidemiol Biomarkers Prev 1997;6:1021–7.
44. Karagas MR, Tosteson TD, Greenberg ER, et al. Effects of milk and milk products on rectal mucosal cell proliferation in humans. Cancer Epidemiol Biomarkers Prev 1998;7:757–66.
45. Cats A, Kleibeuker JH, Van der MR, et al. Randomized, double-blinded, placebo-controlled intervention study with supplemental calcium in families with hereditary nonpolyposis colorectal cancer. J Natl Cancer Inst 1995;87:598–603.
46. Armitage NC, Rooney PS, Gifford KA, Clarke PA, Hardcastle JD. The effect of calcium supplements on rectal mucosal proliferation. Br J Cancer 1995;71:186–90.
47. Thomas MG, Thomson JP, Williamson RC. Oral calcium inhibits rectal epithelial proliferation in familial adenomatous polyposis. Br J Surg 1993;80:499–501.
48. Wargovich MJ , Isbell G, Shabot M, et al. Calcium supplementation decreases rectal epithelial cell proliferation in subjects with sporadic adenoma. Gastroenterology 1992;103:92–7.
49. Lipkin M, Newmark H. Effect of added dietary calcium on colonic epithelial-cell proliferation in subjects at high risk for familial colonic cancer. N Engl J Med 1985;313:1381–4.
50. Stern HS, Gregoire RC, Kashtan H, Stadler J, Bruce RW. Long-term effects of dietary calcium on risk markers for colon cancer in patients with familial polyposis. Surgery 1990;108:528–33.
51. Sandler RS, Baron JA, Tosteson TD, Mandel JS, Haile RW. Rectal mucosal proliferation and risk of colorectal adenomas: results from a randomized controlled trial. Cancer Epidemiol Biomarkers Prev 2000;9:653–6.
52. Willett WC. Diet and cancer: one view at the start of the millennium. Cancer Epidemiol Biomarkers Prev 2001;10:3–8.
53. Flood A, Velie EM, Sinha R, et al. Meat, fat, and their subtypes as risk factors for colorectal cancer in a prospective cohort of women. Am J Epidemiol 2003;158:59–68.
54. Garland CF, Garland FC. Do sunlight and vitamin D reduce the likelihood of colon cancer? Int J Epidemiol 1980;9:227–31.